

AMENDMENT

Amendments to the Claims:

Please amend the claims as follows, without prejudice:

In the Claims:

1. (Previously Presented) A method for amplifying one or more nucleic acids onto a bead comprising the steps of:

(a) forming a water-in-oil emulsion to create a plurality of aqueous microreactors wherein a plurality of the microreactors comprises on average one single stranded nucleic acid template, a single bead with a first population comprising a plurality of molecules of a first primer species disposed thereon, and an amplification reaction solution comprising a second population comprising a plurality of molecules of the first primer species and a plurality of molecules of a second primer species and reagents necessary to perform nucleic acid amplification, wherein the first primer species is capable of binding to the single stranded nucleic acid template, the second primer species is capable of binding to a complementary strand of the single stranded nucleic acid template, and further wherein the molecules of the second primer species and the molecules of the first population of the primer species are each present in greater numbers within the aqueous microreactors than the number of molecules of the second population of the first primer species;

(b) asymmetrically amplifying the single stranded nucleic acid template and the complementary strand to the template strand in the amplification reaction solution to form a population of amplified copies of the single stranded template nucleic acid, wherein substantially all of the molecules of the second population of the first primer species in the amplification reaction solution are depleted;

(c) binding a plurality of the asymmetrically amplified copies of the single stranded template nucleic acid to the first population of the first primer species on the bead in a plurality of the microreactors, wherein at least 100,000 bead bound complementary strands are extended

from the first primer species to form a population of beads with amplified nucleic acid template bound thereto;

(d) breaking the emulsion to release the population of beads with amplified nucleic acid template bound thereto from the microreactors and away from the amplification reaction solution comprising unbound amplification products; and

(e) enriching for beads with amplified nucleic acid template bound thereto by removing beads to which no nucleic acid is bound; and

(f) distributing the beads with amplified nucleic acid template bound thereto onto an array.

2. (Previously Presented) The method of claim 1, wherein a majority of the microreactors include a single nucleic acid.

3. (Previously Presented) The method of claim 1, wherein said amplification reaction solution is a polymerase chain reaction solution further comprising nucleotide triphosphates, a thermostable polymerase, and a buffer compatible with polymerase chain reaction conditions.

4. (Cancelled).

5. (Cancelled).

6. (Previously Presented) The method of claim 1, wherein said emulsion additionally contains emulsion stabilizers.

7. (Previously Presented) The method of claim 6, wherein said emulsion stabilizers are selected from the group consisting of Atlox 4912, Span 80, and combinations and mixtures thereof.

8. (Previously Presented) The method of claim 1 wherein said emulsion is heat stable.

9. (Previously Presented) The method of claim 8 wherein said emulsion is heat stable to 95°C.

10. (Previously Presented) The method of claim 1, wherein amplification is carried out by a method selected from the group consisting of transcription-based amplification, rapid amplification of cDNA ends, continuous flow amplification, and rolling circle amplification.

11. (Previously Presented) The method of claim 1, wherein the emulsion is formed by the dropwise addition of the nucleic acid templates, beads, and amplification reaction solution into an oil.

12. (Previously Presented) The method of claim 1, performed with at least 10,000 nucleic acid templates.

13. (Previously Presented) The method of claim 1, performed with at least 50,000 nucleic acid templates.

14. (Previously Presented) The method of claim 1, wherein the microreactors have an average size of 50 μm .

15. (Previously Presented) The method of claim 1, wherein each bead binds more than 10,000 asymmetrically amplified copies of the single stranded nucleic acid template.

16. to 21. (Cancelled)

22. (Previously Presented) The method of claim 1 further comprising the step of:

(g) sequencing the amplified nucleic acid templates.

23. (Cancelled)

24. (Cancelled).

25. (Previously Presented) The method of claim 1 wherein enrichment step (f) is performed using magnetic beads having primers attached thereto that bind the amplified nucleic acid template.

26. (Cancelled)

27. (Previously Presented) The method of claim 1, wherein at least 1,000,000 copies of each target nucleic acid molecule are bound to each bead.
28. (Previously Presented) The method of claim 1, wherein between at least 1 to 20,000,000 copies of each target nucleic acid molecule are bound to each bead.
29. (Previously Presented) The method of claim 1, wherein the beads are sepharose beads.
30. (Cancelled).
31. (Cancelled).
32. (Previously Presented) The method of claim 25, further comprising, after step (e), the step of: separating the beads with amplified nucleic acid template thereon away from the magnetic beads.
33. (Currently Amended) The method of claim 32, wherein the separating is achieved by incubation at a temperature greater than 45°C or by incubating the beads with amplified nucleic acid template thereon and the magnetic beads in a solution with a basic pH.
34. to 44. (Cancelled).